

monary granuloma formation in murine toxocariasis: transfer of granulomatous hypersensitivity using bronchoalveolar lavage cells. *Journal of Parasitology* 74:950-956.

\_\_\_\_\_, \_\_\_\_\_, and \_\_\_\_\_. 1987. Use of bronchoalveolar lavage to compare local pulmonary immunity with the systemic immune response of *Toxocara canis*-infected mice. *Infection and Immunity* 55:2132-2136.

\_\_\_\_\_, P. E. Omholt, and R. B. Grieve. 1985. Immune responses of CBA/J mice to graded infections with *Toxocara canis* infective larvae. *Infection and Immunity* 48:697-703.

Kazura, J. W., and M. Aikawa. 1980. Host defense mechanisms against *Trichinella spiralis* infection in the mouse; eosinophil mediated destruction of new-born larvae in vitro. *Journal of Immunology* 124:355-361.

\_\_\_\_\_, and D. I. Grove. 1978. Stage-specific antibody dependent eosinophil-mediated destruction of *Trichinella spiralis*. *Nature* 274:355-361.

McLaren, D. J., and F. J. Ramalho-Pinto. 1979. Eosinophil-mediated killing of schistosomula of *Schistosoma mansoni* in vitro: synergistic effect of antibody and complement. *Journal of Immunology* 123:1431-1438.

Ouissi, M. A., A. Haque, and A. Capron. 1981. *Dipetalonema viteae*. Ultrastructural study of the in vitro interaction between rat macrophages and microfilariae in the presence of IgE antibody. *Parasitology* 82:55-68.

Ramalho-Pinto, F. J., D. J. McLaren, and S. R. Smithers. 1978. Complement-mediated killing of schistosomula of *Schistosoma mansoni* by rat eosinophils in vitro. *Journal of Experimental Medicine* 147:147-156.

Sakanari, J. A., H. M. Loinaz, T. L. Deardorff, R. B. Raybourne, J. H. McKerrow, and J. G. Frierson. 1988. Intestinal anisakiasis: a case diagnosed by morphologic and immunologic methods. *American Journal of Clinical Pathology* 90:107-113.

Vadas, M. A., A. E. Butterworth, B. Sherry, A. Dessein, M. Hogan, D. Bout, and J. R. David. 1980. Interactions between human eosinophils and schistosomula of *Schistosoma mansoni*. I. Stable and irreversible antibody-dependent adherence. *Journal of Immunology* 124:1441-1448.

Yen, P. K. F., P. G. Holt, N. F. Stanley, J. M. Papadimitriou, and T. Robertson. 1986a. In vitro antibody-mediated macrophage activity on *Breinlia macropi* microfilariae. II. Ultrastructural and video recording investigations of adherence and cytotoxicity. *Parasite Immunology* 8:201-216.

\_\_\_\_\_, \_\_\_\_\_, \_\_\_\_\_, and K. J. Turner. 1986b. In vitro antibody-mediated macrophage activity on *Breinlia macropi* microfilariae. I. Adherence and cytotoxicity. *Parasite Immunology* 8:139-147.

J. Helminthol. Soc. Wash.  
58(1), 1991, pp. 137-140

### Research Note

## *Sphyranura euryceae* (Monogenea) on *Eurycea* spp. (Amphibia: Caudata), from Northcentral Arkansas

CHRIS T. MCALLISTER,<sup>1</sup> STANLEY E. TRAUTH,<sup>2</sup> AND LAWRENCE W. HINCK<sup>2</sup>

<sup>1</sup> Renal-Metabolic Laboratory (151-G), Department of Veterans Affairs Medical Center,  
4500 S. Lancaster Road, Dallas, Texas 75216 and

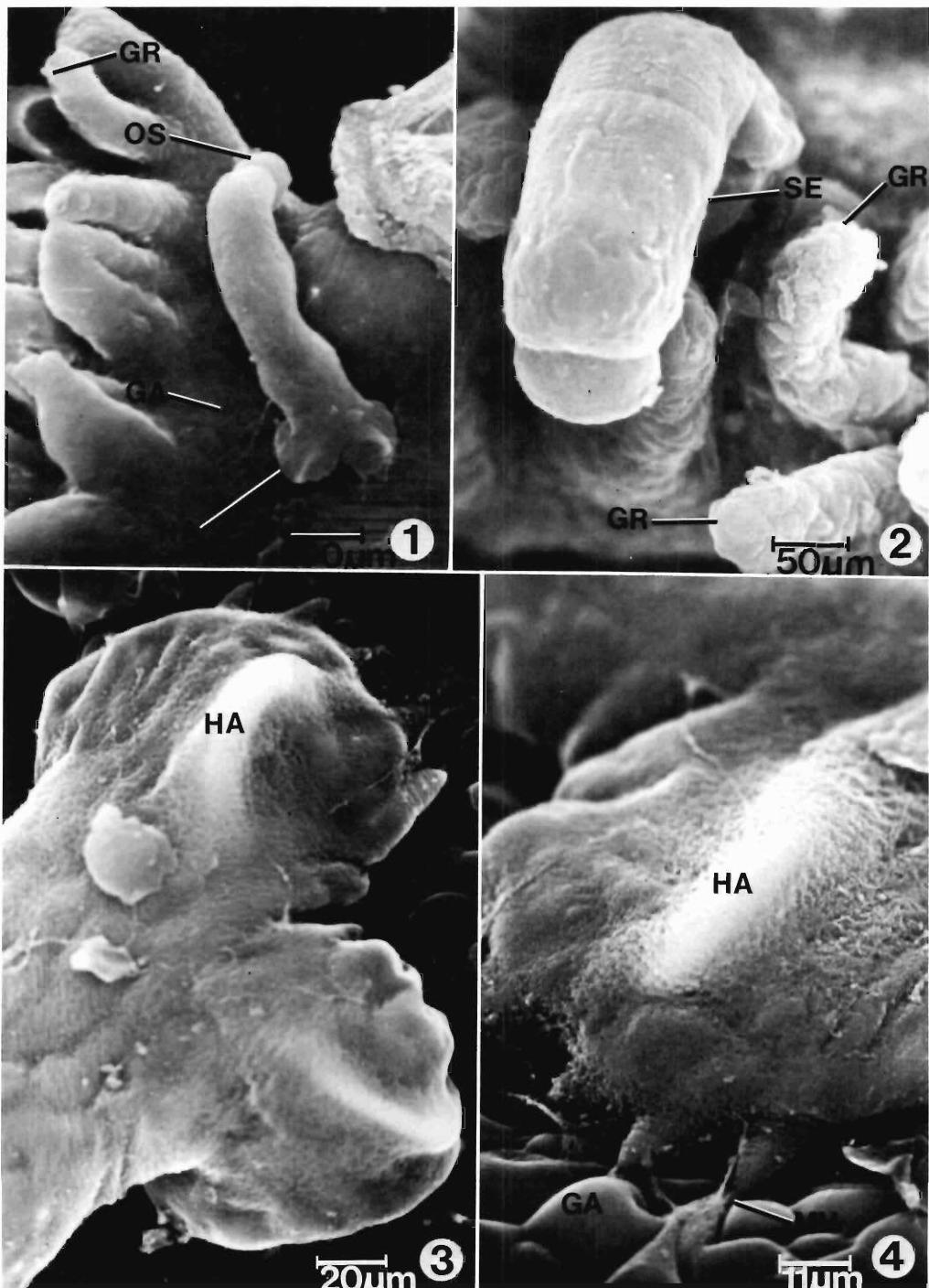
<sup>2</sup> Department of Biological Sciences, Arkansas State University, State University, Arkansas 72467

**ABSTRACT:** *Sphyranura euryceae* Hughes and Moore, 1943 (Polystomatoidea: Sphyranuridae) was found on the external gills, skin, and gular folds of 10/10 larval cave salamanders, *Eurycea lucifuga*, and 10/10 neotenic graybelly salamanders, *E. multiplicata griseogaster*, in northcentral Arkansas. This represents the second time *S. euryceae* has been reported and constitutes new host and distributional records for the parasite. A summary is presented on the *Sphyranura* spp. from caudate amphibians. Based on morphological similarities and the opinions of earlier workers, the synonymy of *S. polyorchis* Alvey, 1936, with *S. osleri* Wright, 1879, is provisionally supported.

**KEY WORDS:** cave salamander, *Eurycea lucifuga*, *E. multiplicata griseogaster*, graybelly salamander, Monogenea, Polystomatoidea, prevalence, *Sphyranura eu-*

*ryceae*, *S. oligorchis*, *S. osleri*, *S. polyorchis*, Sphyranuridae, synonymy.

Wright (1879) described *Sphyranura osleri* from the gills and mouth cavity of mudpuppies, *Necturus lateralis* (syn. of *N. maculosus*). Wright and Macallum (1887) provided additional information about the species. Since then, additional *Sphyranura* spp. have been described or reported from North American hosts (Alvey, 1933a, b, 1936; Hughes and Moore, 1943; Coggins and Sajdak, 1982). The purpose of this note is to report *Sphyranura euryceae* Hughes and



Figures 1–4. Scanning electron micrographs of *Sphyranura euryceae* infesting neotenic *Eurycea multiplicata griseogaster* from Arkansas. 1. Worm attached to gill arch (GA) and gill rakers (GR) showing oral sucker (OS) and haptor (HA). 2. Closer view of worm (SE) on gill rakers (GR). 3. Attachment of haptor (HA) on gill arch. 4. Higher magnification of haptor (HA); note marginal hook (MH) attached to gill arch (GA).

**Table 1.** Species of *Sphyranura* reported from North American Caudata.

<i>Sphyranura</i> spp.	Host(s)	Localities	Reference(s)
<i>S. euryceae</i>	<i>Eurycea tynerensis</i> ; <i>E. multiplicata griseogaster</i> , <i>E. lucifuga</i>	Oklahoma; Arkansas	Hughes and Moore, 1943; McAllister et al., this report
<i>S. oligorchis</i>	<i>Necturus maculosus</i>	Pennsylvania	Alvey, 1933a, b, 1936
<i>S. osleri</i>	<i>N. maculosus</i>	Canada; Wisconsin	Wright, 1879; Wright and Macallum, 1887; Coggins and Sajdak, 1982
<i>S. polyorchis*</i>	<i>N. maculosus</i>	Pennsylvania	Alvey, 1936

\* May be a synonym of *S. osleri*.

Moore, 1943, on 2 species of plethodontid salamanders from Arkansas and to comment on possible synonymy of *S. polyorchis* Alvey, 1936.

During January 1990, 10 larval cave salamanders, *Eurycea lucifuga* Rafinesque, 1822, and 10 neotenic graybelly salamanders, *E. multiplicata griseogaster* Moore and Hughes, 1941, were collected 2.4 km W Lakeway off Hwy 14 at Chapman Spring, Marion County. Salamanders were captured by hand, placed in individual plastic bags, and on return to the laboratory (within 24 hr), were killed by immersion in a dilute chloretoe solution, fixed in 10% formalin, and stored in 70% ethanol. Monogeneans were noted on all salamanders and some were teased from gill filaments, gular folds, or skin to be flattened under gentle cover glass pressure. Specimens were later stained with acetocarmine, dehydrated through an ethanol series, cleared in xylene, and mounted in Permount®. Techniques used to prepare tissues of other infested salamanders for SEM followed routine procedures and included dehydration in an ethanol series with amyl acetate transition solvent. Specimens were dried in a Samdri critical point dryer and coated with gold/palladium in a Hummer IV sputter coater from 2 to 5 min. A JEOL 100 CXII TEM-SCAN electron microscope was used to view gills and skin at an accelerating voltage of 20 kV. Representative specimens of *S. euryceae* have been deposited in the USNM Helminthological Collection, USDA, Beltsville, Maryland 20705, as USNM 81047. Voucher specimens of salamanders have been deposited in the Arkansas State University Museum of Zoology (ASUMZ) as follows: *E. lucifuga* (ASUMZ 15348–15357) and *E. m. griseogaster* (ASUMZ 15338–15347, 15358–15359).

Of the 20 salamanders examined, all were harboring at least 1 polystomatid monogenean identified as *Sphyranura euryceae* (Hughes and

Moore, 1943). Numerous worms were observed attached to gill arches and rakers of individual salamanders and are shown in Figures 1–4. Five *S. euryceae* had the following characteristics and measurements (mean followed by the range in micrometers [ $\mu\text{m}$ ] in parentheses): total body length 1,620 (800–2,400); maximum width 420 (300–600); haptor 463 (269–767) wide by 259 (191–355) in maximum length; caudal sucker diameter 137 (111–197); oral sucker width 203 (155–284) by 186 (153–225) long ( $N = 4$ ); spheroid to subspheroidal testes 7 (4–8) in number, 102 (78–120) wide by 77 (53–98) long ( $N = 6$ ); uterus obscured in 2 specimens and occupied by a single, heavy-shelled, egg 190 (145–217) wide by 268 (254–282) long ( $N = 3$ ).

Hughes and Moore (1943) reported *S. euryceae* was found on 45 of 90 Oklahoma salamanders, *Eurycea tynerensis* Moore and Hughes, 1939, from Cherokee County, Oklahoma. The type locality of *S. euryceae* (Pea Vine Creek near Tahlequah) is approximately 225 km WSW of the locality reported herein.

Although there have been several studies on the helminth parasites of *Eurycea lucifuga* involving 370 salamanders (Landewe, 1963; Dyer and Brandon, 1973; Dyer and Peck, 1975; Castle et al., 1987), *Sphyranura* spp. was not reported. Perhaps this is a consequence of surveying only metamorphosed terrestrial adults, since species of *Sphyranura* have been reported previously to infest only aquatic larval or neotenic salamanders having external gills (Table 1).

Four species of *Sphyranura* are known from North American caudate amphibians (Table 1). However, the validity of *S. polyorchis* Alvey, 1936, was questioned by Price (1939) who noted that the subtle differences between *S. osleri* and *S. polyorchis*, claimed by Alvey (1936), probably do not justify the recognition of 2 separate species. This view was later reiterated by Hughes

and Moore (1943). According to Price (1939) there are no specimens of *S. polyorchis* available and except for differences in number of testes (20–23 in *S. polyorchis* and 12–16 in *S. osleri*) and in the supposed absence of spines on large haptoral hooks of *S. polyorchis*, the 2 species are essentially the same in all other characteristics and measurements. Compelling evidence for recognizing distinct species is not evident and it is possible that the differences noted by Alvey (1936) may be due to individual variation. Therefore, the synonymy originally proposed by Price (1939) is provisionally supported until specimens of *S. polyorchis* can be rediscovered and examined.

S.E.T. thanks the Arkansas Game and Fish Commission for Scientific Collecting Permit No. 831.

#### Literature Cited

**Alvey, C. H.** 1933a. *Sphyranura oligorchis* n. sp. from *Necturus maculosus*. *Journal of Parasitology* 20: 140.

—. 1933b. The life cycle of *Sphyranura oligorchis*. *Journal of Parasitology* 20:140.

—. 1936. The morphology and development of the monogenetic trematode *Sphyranura oligorchis* (Alvey, 1933) and the description of *Sphyranura polyorchis* n. sp. *Parasitology* 28:229–259.

**Castle, M. D., D. A. Strohlein, and B. M. Christensen.** 1987. Helminth parasites of the cave salamander, *Eurycea lucifuga*, from western Kentucky. *Proceedings of the Helminthological Society of Washington* 54:269–270.

**Coggins, J. R., and R. A. Sajdak.** 1982. A survey of helminth parasites in the salamanders and certain anurans from Wisconsin. *Proceedings of the Helminthological Society of Washington* 49:99–102.

**Dyer, W. G., and R. A. Brandon.** 1973. Helminths of three sympatric species of cave-dwelling salamanders in southern Illinois. *Transactions of the Illinois State Academy of Science* 66:23–29.

—, and S. B. Peck. 1975. Gastrointestinal parasites of the cave salamander, *Eurycea lucifuga* Rafinesque, from the southeastern United States. *Canadian Journal of Zoology* 53:52–54.

**Hughes, R. C., and G. A. Moore.** 1973. *Sphyranura euryceae*, a new polystomatid monogenean fluke from *Eurycea tynerensis*. *Transactions of the American Microscopical Society* 62:286–292.

**Landewe, J. E.** 1963. Helminth and arthropod parasites of salamanders from southern Illinois. Unpubl. M.S. Thesis, Southern Illinois University, Carbondale. 47 pp.

**Price, E. W.** 1939. North American monogenetic trematodes. IV. The family Polystomatidae (Polystomatoidea). *Proceedings of the Helminthological Society of Washington* 6:80–92.

**Wright, R. R.** 1879. Contributions to American helminthology. No. I. *Proceedings of the Canadian Institute, Toronto* 1:54–75.

—, and A. B. Macallum. 1887. *Sphyranura osleri*: a contribution to American helminthology. *Journal of Morphology* 1:1–48.

J. Helminthol. Soc. Wash.  
58(1), 1991, pp. 140–142

#### Research Note

### Long-term Storage of Hookworm Infective Larvae in Buffered Saline Solution Maintains Larval Responsiveness to Host Signals

J. M. HAWDON AND G. A. SCHAD

Department of Pathobiology, University of Pennsylvania, Philadelphia, Pennsylvania 19104

**ABSTRACT:** Third-stage larvae ( $L_3$ 's) of *Ancylostoma caninum* stored in water exhibited a decline in the number of larvae that resumed feeding in response to canine serum, whereas those stored in copro-culture for the same amount of time failed to show this decline. When  $L_3$ 's were stored for 39 days in BU, a *Caenorhabditis elegans* handling buffer, they retained the ability to resume feeding. Short (<24 hr) storage in water had no effect on feeding.

**KEY WORDS:** *Ancylostoma caninum*, host signals, hookworm, infective larvae.

During investigations of the resumption of feeding by infective hookworm larvae ( $L_3$ 's) when exposed to host-mimicking conditions *in vitro* (Hawdon and Schad, 1990), we have observed a marked decrease in the proportion of larvae responding to a feeding stimulus when the larvae were first stored in water. Larvae that remained in copro-culture for the same length of time failed to exhibit this decline in the proportion feeding,